

Pre Edit

*RETURN TO FMF - LOCATION 7540

QUERY CONTROL FORM		RTIS USE ONLY	
Application No.	091057289	Prepared by	BRS
Examiner-GAU	Dobodansky 1652	Date	1-27-04
		No. of queries	1
X			

JACKET			
a. Serial No.	f. Foreign Priority	k. Print Claim(s)	p. PTO-1449
b. Applicant(s)	g. Disclaimer	l. Print Fig.	q. PTOL-85b
c. Continuing Data	h. Microfiche Appendix	m. Searched Column	r. Abstract
d. PCT	i. Title	n. PTO-270/328	s. Sheets/Figs
e. Domestic Priority	j. Claims Allowed	o. PTO-892	t. Other

SPECIFICATION a. Page Missing b. Text Continuity c. Holes through Data d. Other Missing Text e. Illegible Text f. Duplicate Text g. Brief Description h. Sequence Listing i. Appendix j. Amendments k. Other <i>Continuing Data Paragraph</i>	MESSAGE <i>Continuing Data: There is no Continuing Data paragraph for claim of benefits for all US provisional application. Please provide</i>		
	<i>Thanks</i>		
CLAIMS a. Claim(s) Missing b. Improper Dependency c. Duplicate Numbers d. Incorrect Numbering e. Index Disagrees f. Punctuation g. Amendments h. Bracketing i. Missing Text j. Duplicate Text k. Other	initials <i>BRS</i>		
	RESPONSE <i>Provisional data inserted on p. 1.</i>		
	<i>Note: Contractor authorized to insert this information on p. 1 when supported by other papers in file.</i>		
	initials <i>JBH</i>		

JBT
2-2-04**LUCIFERASE EXPRESSION CASSETTES AND METHODS OF USE**

This application claims the benefit of provisional application No. 60/152904, filed Sept. 8, 1999.

TECHNICAL FIELD

The present invention relates to luciferase expression vectors, methods of making same and methods of use thereof.

BACKGROUND OF THE INVENTION

Bioluminescent bacteria are widely found in both marine and terrestrial environments. Interestingly, all identified species of naturally occurring marine and terrestrial bioluminescent bacteria are Gram-negative. To date, at least eleven species in four Gram-negative genera have been described: *Vibrio*, *Photobacterium*, *Shewanella (Altermonas)* and *Photorhabdus (Xenorhabdus)*. In all these species, the five genes responsible for bioluminescence are clustered in the *lux* operon (*luxCDABE*).

The bioluminescence (emitted blue-green light having a wavelength of about 490 nm) is thought to result from a luciferase-catalyzed oxidation of reduced flavin mononucleotide (FMNH₂) and a long-chain fatty aldehyde. The luciferase enzyme is encoded by two subunits (*luxAB*), whereas the fatty acid reductase polypeptides responsible for the biosynthesis of the aldehyde substrate for the luminescent reaction are encoded by the three genes *luxCDE*. The genes encoding luciferase and the fatty acid reductase polypeptides have been cloned from the *lux* operons of *Vibrio*, *Photobacterium* and *Photorhabdus* and sequenced. In each case, the *luxCDE* genes flank the *luxAB* genes, with transcription in the order *luxCDABE*. Although a number of additional *lux* genes have been identified in each of these three bacteria, only *luxA-E* are essential for the biosynthesis of light (reviewed by Meighen, E., (1993, *The FASEB Journal* 7:1016-1022 and Ülitzur, S., (1997, *J. Biolumin Chemilumin* 12:179-192).

Methods described in U.S. Patent 5,650,135, make possible the detection of bioluminescent bacteria in a living animal without dissecting or otherwise opening the animal up ("in vivo monitoring") – the light is detected through muscle, skin, fur & other traditionally "opaque" tissues using a highly sensitive camera. In this context and others, it would therefore be desirable to confer bioluminescence properties on a bacterium of one's choice, so that the bacterium could be followed with *in vivo* monitoring in various models of infection. In particular, it would be desirable to confer such bioluminescence properties on Gram-positive